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# ON THE HYDROGEN ION CONCENTRATION OF SEA WATER, AND THE PHYSIOLOGICAL EFFECTS OF THE IONS OF SEA WATER

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Both natural sea water at Tortugas, Florida, of salinity of 35–36 per kilo and artificial sea water of the following composition were studied:

	<i>Normal solutions</i>		or	<i>Isotonic solutions.</i>	
CaCl <sub>2</sub> .....	0.5 m.	22.07 cc.		0.38 m.	29.0 cc.
MgCl <sub>2</sub> .....	0.5 m.	50.21 cc.		0.37 m.	67.9 cc.
MgSO <sub>4</sub> .....	0.5 m.	57.09 cc.		0.975 m.	29.5 cc.
KCl.....	1.0 m.	10.23 cc.		0.577 m.	17.7 cc.
NaCl.....	1.0 m.	483.65 cc.		0.568 m.	852.0 cc.
NaBr.....	1.0 m.	0.80 cc.		0.565 m.	1.4 cc.
NaHCO <sub>3</sub> .....	1.0 m.	2.40 cc.		0.93 m.	2.58 cc.
H <sub>2</sub> O.....		373.63 cc.			0.00 cc.
		1000.00 cc.			1000.00 cc.

The mixture was aerated many hours or until it reached the proper alkalinity. It was tested by placing the most delicate marine organisms in it, which were found to live and behave normally.

The excess of non-volatile base over mineral acid in Tortugas sea water was found by Dole to be 0.002265–0.00253 N. per kilogram, and my own observations fall within these limits. That of various artificial sea waters studied was 0.00225–0.00245 Normal per kilogram. This variation in natural sea water is independent of the salinity and is apparently associated with the variation in the lime content.

The hydrogen ion concentration will be indicated by the minus logarithm or PH, the PH of pure water, or neutral solution, at 23° being 7, that of *n.* HCl ca. 0, and that of *n.* KOH ca. 14. The PH was determined by the hydrogen electrode and standardized instruments, the special apparatus being described by McClendon and Magoon<sup>1</sup> All measurements were made at 30°. The PH of Tortugas sea water was found to vary from 8.1 to 8.22 and this variation was found to be partly due to the variation in the excess base but largely due to variations in the CO<sub>2</sub> tension.

The PH of sea water at known CO<sub>2</sub> tensions was studied and it was found that, as the CO<sub>2</sub> tension decreased, the ratio of change in PH with change in CO<sub>2</sub> tension increased so that the PH could be much more accurately measured than the CO<sub>2</sub> tension. The CO<sub>2</sub> tension of the

sea varied from about 0.03–0.05% of an atmosphere, but these measurements will be made more accurately next summer. One estimation of the  $\text{CO}_2$  of the air was 0.033%. It seems probable that  $\text{CO}_2$  is passing from the sea to the air at Tortugas, but this question also will be studied more accurately next summer. From Fox's data and the above measurements, the following estimations at  $30^\circ$  may be made:

$\text{CO}_2$ content in cc. per litre of sea water .....	44.5	46.25	47.5
$\text{CO}_2$ tension in percent of an atmosphere.....	0.03	0.04	0.05
PH.....	8.22	8.15	8.1

From this table the  $\text{CO}_2$  content may be estimated from the PH and hence in respiration may be made by estimating the PH of water containing animals, at certain periods. For this reason rapid methods of estimating the PH were calibrated by comparing vials of Sørensen's borate mixtures colored with thymolsulphonephthalein<sup>2</sup> with a similar vial of sea water to which is added the same quantity of thymolsulphonephthalein, and the PH read off colorimetrically. It was found that when the sea water was tested with the same hydrogen electrode used in standardizing the borate mixtures its PH was found to be 0.3 lower than that estimated colorimetrically, owing to the salt action on the indicator, consequently 0.3 must be subtracted from the result obtained colorimetrically. The salt error could not be determined more accurately with these tubes since they read down only to 0.1 on the PH scale but I understand that Lubs and Clark are studying this question in detail and may have more exact data.

Some general experiments on the physiological significance of the ions of sea water were made, but since they were intended to be general, no extremely exact quantitative data were recorded. Hence these experiments cannot be compared with those of A. G. Mayer on the same subject in which he has made some exact determinations of the effect of H and OH ions at various concentrations. Since I have shown that ions may change the permeability of irritable cells, and excitation seems evidently to be associated with increase in permeability to ions and I have also shown that it is accompanied by increase in electrical conductivity; the question arises whether all ions may be divided into two classes: those which increase permeability and those which inhibit this change. Experiments on the pulsation of the jelly fish, *Cassiopea*, and the heart of the Conch, *Strombus*, indicate that  $\text{OH}'$ ,  $\text{Na}'$ , and  $\text{K}'$  increase permeability, and that  $\text{H}'$ ,  $\text{Mg}'$ , and  $\text{Ca}'$  inhibit increase in permeability. Since a certain increase in permeability puts the cell in the stimulated condition which is the same as the re-

fractory period,  $K^+$  may inhibit stimulation by giving the cell a single stimulus which continues and is manifested by the continuance of the refractory period as long as the potassium ion is present.  $Ca^{++}$  on the other hand is very toxic and when too liberally applied causes death accompanied by increase in permeability and death rigor, which may simulate the stimulated state.

$OH^+$ ,  $Na^+$ , and  $K^+$  lower the threshold for excitation of the jellyfish and conch heart, and  $H^+$ ,  $Mg^{++}$  and  $C^{++}$  raise it. When cautiously applied,  $OH^+$ ,  $Na^+$ , and  $K^+$  stop the conch heart in systole, and  $H^+$ ,  $Mg^{++}$ , and  $Ca^{++}$  stop it in diastole, but the action of  $Ca^{++}$  may seem the reverse when not carefully studied. This seemingly uncertain condition is probably due to the organism being composed of several kinds of irritable elements, each requiring a balance of ions of the two classes. A balance for all these elements cannot be maintained with one pair of antagonistic ions alone.

Since the plasma membrane or cell surface is very probably composed of emulsoids, or hydrophile colloids,<sup>3</sup> the change in permeability must be due to a colloid change. Although suspensoids are easily coagulated or precipitated, emulsoids have too great an affinity for water to be easily separated from it except in a most gradual manner unless they are changed by certain reagents or temperatures so as to approach the character of suspensoids. We should therefore not look toward the processes of aggregation and dispersion as seen in suspensoids for an explanation of change in permeability. The changes in an emulsoid membrane are to be regarded as swelling and shrinkage as seen in gelatine or collodion plates. Water soluble substances diffuse easily through collodion gels until the shrinkage reaches a certain point, beyond which the permeability rapidly decreases with shrinkage and the collodion finally becomes impermeable to electrolytes. This is probably due to the approximation of the collodion molecules until the pores are too small to admit of free diffusion of hydrated ions through them. It is to be distinguished from mere approximation of colloid particles, since we may have diffusion through a continuous sheet that has swelled in water, and the diffusion probably takes place in the bound or hydrate water itself. Bartell and Bigelow have shown that porcelain becomes rapidly semipermeable when the pores decrease below 0.1 micron.

It was shown by H. N. Morse that KCl and NaCl increase the permeability of colloidal copper ferrocyanide membranes, but it is a generally known fact that Ca and Mg salts do not increase the permeability, at least not sufficiently for their own diffusion or of that of sugar. Na and K salts (especially chlorides) increase the swelling and permeability

of gelatine plates and Mg and Ca salts, at least under certain conditions, retard the swelling and may even cause shrinkage.  $\text{OH}'$  increases swelling and  $\text{H}'$  decreases swelling if the reaction of the medium is on the alkaline side of the isoelectric point for the colloid, which seems to be the case in regard to blood and animal proteins within the body or in sea water.

We may therefore conclude that  $\text{OH}'$ ,  $\text{Na}'$ , and  $\text{K}'$  increase the permeability of the plasma membrane by causing it to swell and that  $\text{Ca}'$ ,  $\text{Mg}'$ , and  $\text{H}'$  (at least on the alkaline side of the isoelectric point) inhibit increase in permeability by inhibiting swelling.

<sup>1</sup> *J. Biol. Chem.*, 25, 669 (1916).

<sup>2</sup> These vials were obtained from Hynson, Westcott and Company, Baltimore, Maryland.

<sup>3</sup> There is reason to support the view that the cell surface is composed of emulsoids rather than suspensoids.

## SOME INTERRELATIONS BETWEEN DIET, GROWTH, AND THE CHEMICAL COMPOSITION OF THE BODY

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Changes which normally occur in the water, ether extract, and ash content of the body during its most active growth have been determined for the white mouse. Based on eighty-eight analyses of the entire body at different stages of growth, the following results were obtained: (a) increase in solids from 16% at birth to a maximum of 35% at fifty days with a subsequent decrease to 33%; (b) decrease in the proportion of water in the fat-free substance from 85.5% at birth to 73% in the adult mouse; (c) rapid increase in fat during the first twelve days from 1.85% to about 10%, followed by a slow increase to about 12%; (d) absolute and relative increase in ash content from 25 mg., 1.86%, in a mouse weighing one and a half grams at birth, to 950 mg., 3%, of ash in the adult mouse.

The ash content of mice growing normally on an artificial food mixture of milk powder, casein, starch, salts, and butter fat, (protein 30%, fat 32%, and ash 5.5%), is uniformly higher at corresponding stages of growth than the ash content of mice fed on a ration of mixed grain, dog bread, and small amounts of milk. It is not known to what this difference is due, but it is not related to differences in the amount of protein or salts in the food. Since the ash content of normal animals may be thus increased by differences in diet alone, it would seem that